

Table 1. Sequence of LoxP Sites

LoxP WT 5'-ATAACTTCGTATAATGTATGCTATACGAAGTTAT-3'
[SEQ ID NO: 1]

LoxP511 5'-ATAACTTCGTATAGTATACATTATACGAAGTTAT-3'
[SEQ ID NO: 2]

LoxC2 5'-ACAACTTCGTATAATGTATGCTATACGAAGTTAT-3'
[SEQ ID NO: 3]

LoxP1 5'-ATAACTTCGTATAATATATGCTATACGAAGTTAT-3'
[SEQ ID NO: 4]

LoxP2 5'-ATAACTTCGTATAGCATAACATTATACGAAGTTAT-3'
[SEQ ID NO: 5]

LoxP3 5'-ATAACTTCGTATAATGTATACTATACGAAGTTAT-3'
[SEQ ID NO: 6]

LoxP4 5'-ATAACTTCGTATAATATAAACTATACGAAGTTAT-3'
[SEQ ID NO: 7]

LoxP5 5'-ATAACTTCGTATAATCTAACCTATACGAAGTTAT-3'
[SEQ ID NO: 8]

LoxP6 5'-ATAACTTCGTATAACATAGCCTATACGAAGTTAT-3'
[SEQ ID NO: 9]

LoxP7 5'-ATAACTTCGTATAACATACCCTATACGAAGTTAT-3'
[SEQ ID NO: 10]

LoxP8 5'-ATTACCTCGTATAGCATAACATTATACGAAGTTAT-3'
[SEQ ID NO: 11]

LoxP9 5'-ATAACTTCGTATAGCATAACATTATATGAAGTTAT-3'
[SEQ ID NO: 12]

LoxP10 5'-ATTACCTCGTATAGCATAACATTATATGAAGTTAT-3'
[SEQ ID NO: 13]

Table 2. Sequence of PCR primers for amplifying heavy- and light-chain genes of human antibody.

(B= C/G/T; D= A/G/T; K= G/T; M= A/C; R= A/G; S= C/G; W= A/T; and Y= C/T)

a) Heavy-chain

5'-primers for Fv:

VH5'1: 5'-ACC AAG GAA AAA CAA GCG GCC GCA CAG GTG CAG CTG
CAG GAG TCS G-3' [SEQ ID NO: 14]

VH5'2: 5'-ACC AAG GAA AAA CAA GCG GCC GCA CAG GTA CAG CTG
CAG CAG TCA-3' [SEQ ID NO: 15]

VH5'3: 5'-ACC AAG GAA AAA CAA GCG GCC GCA CAG GTG CAG CTA
CAG CAG TGG G-3' [SEQ ID NO: 16]

VH5'4: 5'-ACC AAG GAA AAA CAA GCG GCC GCA GAG GTG CAG CTG
KTG GAG WCY-3' [SEQ ID NO: 17]

VH5'5: 5'-ACC AAG GAA AAA CAA GCG GCC GCA CAG GTC CAG CTK
GTR CAG TCT GG-3' [SEQ ID NO: 18]

VH5'6: 5'-ACC AAG GAA AAA CAA GCG GCC GCA CAG RTC ACC TTG
AAG GAG TCT G-3' [SEQ ID NO: 19]

VH5'7: 5'-ACC AAG GAA AAA CAA GCG GCC GCA CAG GTG CAG CTG
GTG SAR TCT GG-3' [SEQ ID NO: 20]

3'-primers for Fv:

VH3'1: 5'-ATC CAC CGC GGT CGA CTA TGA GGA GAC RGT GAC CAG
GGT G-3' [SEQ ID NO: 21]

VH3'2: 5'-ATC CAC CGC GGT CGA CTA TGA GGA GAC GGT GAC CAG
GGT T-3' [SEQ ID NO: 22]

VH3'3: 5'-ATC CAC CGC GGT CGA CTA TGA AGA GAC GGT GAC CAT
TGT-3' [SEQ ID NO: 23]

VH3'4: 5'-ATC CAC CGC GGT CGA CTA TGA GGA GAC GGT GAC CGT
GGT CC-3' [SEQ ID NO: 24]

VH3'5: 5'-ATC CAC CGC GGT CGA CTA GGT TGG GGC GGA TGC ACT
CC-3' [SEQ ID NO: 25]

VH3'6: 5'-ATC CAC CGC GGT CGA CTA SGA TGG GCC CTT GGT GGA
RGC-3' [SEQ ID NO: 26]

3'-primer for IgG CH1 region of Fab:

FabH3'1: 5'-ATC CAC CGC GGT CGA CTA ACA TGG TTT GVR CTC
AAC TBT CTT GTC CAC-3' [SEQ ID NO: 27]

3'-primer for IgG CH3 region of Ab:

AbH3'1: 5'-ATC CAC CGC GGT CGA CTA TTT ACC CRG AGA CAG
GGA GAG GCT-3' [SEQ ID NO: 28]

**b) Light-chain V λ for cloning into a site downstream of
GAL-4 AD**

5'-primers for Fv:

V λ 5'1: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT CAG TCT GTS BTG ACG CAG
CCG CC-3' [SEQ ID NO: 29]

V λ 5'2: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT TCC TAT GWG CTG ACW CAG
CCA C-3' [SEQ ID NO: 30]

V λ 5'3: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT TCC TAT GAG CTG AYR CAG
CYA CC-3' [SEQ ID NO: 31]

V λ 5'4: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT CAG CCT GTG CTG ACT CAR
YC-3' [SEQ ID NO: 32]

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Vλ5'5: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT CAG DCT GTG GTG ACY CAG
GAG CC-3' [SEQ ID NO: 33]

Vλ5'6: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT CAG CCW GKG CTG ACT CAG
CCM CC-3' [SEQ ID NO: 34]

Vλ5'7: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT TCC TCT GAG CTG AST CAG
GAS CC-3' [SEQ ID NO: 35]

Vλ5'8: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT CAG TCT GYY CTG AYT CAG
CCT-3' [SEQ ID NO: 36]

Vλ5'9: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT AAT TTT ATG CTG ACT CAG
CCC C-3' [SEQ ID NO: 37]

3'-primers for Fv:

Vλ3'1: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TAG GAC GGT SAS CTT GGT
CC-3' [SEQ ID NO: 38]

Vλ3'2: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC GAG GAC GGT CAG CTG GGT
GC-3' [SEQ ID NO: 39]

3'-primer for Cλ1 region

Abλ3'1: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TTA TGA ACA TTC TGC AGG
GGC MAC TGT-3' [SEQ ID NO: 40]

3'-primer for Cλ2 region

127

Abλ3'2: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TTA AGA GCA TTC TGC AGG
GGC CAC TGT-3' [SEQ ID NO: 41]

c) Light-chain Vk for cloning into a site downstream of
GAL-4 AD

5'-primers for Fv:

Vk5'1: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT GAC ATC CRG DTG ACC CAG
TCT CC-3' [SEQ ID NO: 42]

Vk5'2: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT GAA ATT GTR WTG ACR CAG
TCT CC-3' [SEQ ID NO: 43]

Vk5'3: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT GAT ATT GTG MTG ACB CAG
WCT CC-3' [SEQ ID NO: 44]

Vk5'4: 5'-CCA CCA AAC CCA AAA AAA GAG ATC TGT ATG GCT TAC
CCA TAC GAT GTT CCA GAT TAC GCT GAA ACG ACA CTC ACG CAG
TCT C-3' [SEQ ID NO: 45]

3'-primers for Fv:

Vk3'1: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TTT GAT TTC CAC CTT GGT
CC-3' [SEQ ID NO: 46]

Vk3'2: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TTT GAT CTC CAS CTT GGT
CC-3' [SEQ ID NO: 47]

Vk3'3: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TTT GAT ATC CAC TTT GGT
CC-3' [SEQ ID NO: 48]

Vk3'4: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TTT AAT CTC CAG TCG TGT
CC-3' [SEQ ID NO: 49]

3' primer for Ck:

Abk3'1: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC CTA GCA CTC TCC CCT GTT
GAA GCT-3' [SEQ ID NO: 50]

d) Light-chain Vλ for cloning into a site upstream of
GAL-4 AD

5'-primers for Fv:

Vλ5'1: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC CAG TCT GTS BTG ACG
CAG CCG CC-3' [SEQ ID NO: 51]

Vλ5'2: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC TCC TAT GWG CTG ACW
CAG CCA C-3' [SEQ ID NO: 52]

Vλ5'3: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC TCC TAT GAG CTG AYR
CAG CYA CC-3' [SEQ ID NO: 53]

Vλ5'4: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC CAG CCT GTG CTG ACT
CAR YC-3' [SEQ ID NO: 54]

Vλ5'5: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC CAG DCT GTG GTG ACY
CAG GAG CC-3' [SEQ ID NO: 55]

Vλ5'6: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC CAG CCW GKG CTG ACT
CAG CCM CC-3' [SEQ ID NO: 56]

Vλ5'7: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC TCC TCT GAG CTG AST
CAG GAS CC-3' [SEQ ID NO: 57]

Vλ5'8: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC CAG TCT GYY CTG AYT
CAG CCT-3' [SEQ ID NO: 58]

Vλ5'9: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC AAT TTT ATG CTG ACT
CAG CCC C-3' [SEQ ID NO: 59]

3'-primers for Fv:

Vλ3'1: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC TAG GAC GGT SAS CTT GGT
CC-3' [SEQ ID NO: 60]

Vλ3'2: 5'-GAG ATG GTG CAC GAT GCA CAG TTG AAG TGA ACT TGC
GGG GTT TTT CAG TAT CTA CGA TTC GAG GAC GGT CAG CTG GGT
GC-3' [SEQ ID NO: 61]

3'-primer for Cλ1 region

Abλ3'1: 5'-GTT AGT GAA AGT GAA GGA CAA TGA GCT ATC AGC AAT
ATT CCC ACT TTG ATT AAA ATT GGC TGA ACA TTC TGC AGG GGC
MAC TGT-3' [SEQ ID NO: 62]

3'-primer for Cλ2 region

Abλ3'2: 5'-GTT AGT GAA AGT GAA GGA CAA TGA GCT ATC AGC AAT
ATT CCC ACT TTG ATT AAA ATT GGC AGA GCA TTC TGC AGG GGC
CAC TGT-3' [SEQ ID NO: 63]

c) Light-chain Vκ for cloning into a site upstream of GAL-
4 AD

5'-primers for Fv:

Vk5'1: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC GAC ATC CRG DTG ACC
CAG TCT CC-3' [SEQ ID NO: 64]

Vk5'2: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC GAA ATT GTR WTG ACR
CAG TCT CC-3' [SEQ ID NO: 65]

Vk5'3: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC GAT ATT GTG MTG ACB
CAG WCT CC-3' [SEQ ID NO: 66]

Vk5'4: 5'-GAT AAA GCG GAA TTA ATT CCC GAG CCT CCA AAA AAG
AAG AGA AAG GTC GAA TTG GGT ACC GCC GAA ACG ACA CTC ACG
CAG TCT C-3' [SEQ ID NO: 67]

3'-primers for Fv:

Vk3'1: 5'-GTT AGT GAA AGT GAA GGA CAA TGA GCT ATC AGC AAT
ATT CCC ACT TTG ATT AAA ATT GGC TTT GAT TTC CAC CTT GGT
CC-3' [SEQ ID NO: 68]

Vk3'2: 5'-GTT AGT GAA AGT GAA GGA CAA TGA GCT ATC AGC AAT
ATT CCC ACT TTG ATT AAA ATT GGC TTT GAT CTC CAS CTT GGT
CC-3' [SEQ ID NO: 69]

Vk3'3: 5'-GTT AGT GAA AGT GAA GGA CAA TGA GCT ATC AGC AAT
ATT CCC ACT TTG ATT AAA ATT GGC TTT GAT ATC CAC TTT GGT
CC-3' [SEQ ID NO: 70]

Vk3'4: 5'-GTT AGT GAA AGT GAA GGA CAA TGA GCT ATC AGC AAT
ATT CCC ACT TTG ATT AAA ATT GGC TTT AAT CTC CAG TCG TGT
CC-3' [SEQ ID NO: 71]

3' primer for Ck:

Abk3'1: 5'-GTT AGT GAA AGT GAA GGA CAA TGA GCT ATC AGC AAT
ATT CCC ACT TTG ATT AAA ATT GGC GCA CTC TCC CCT GTT GAA
GCT-3' [SEQ ID NO: 72]

Table 3. Sequence of oligonucleotides for modifying the cloning vector pACT2.

a) Oligos for modifying the original MCS in pBridge:

Sac2 Pvu2 Sal I

Sequence A1 5'-pGATCCGCGGCAGCTGTCGAC-3'

[SEQ ID NO. 73]

Sequence A2 5'-pGTACGTCGACAGCTGCCGCG-3'

[SEQ ID NO. 74]

b) Oligos for amplifying the P_{met25} expression cassette in pBridge:

Sequence A3: oligo corresponding to the 5' end of (P_{MET25})

[SEQ ID NO: 75]

Xho I

5'-ACTCGAGCTTCTAATTCTTCCAACATAC

Sequence A4: oligo complementing to the 3' end of (T_{PGK})

[SEQ ID NO: 76]

Xho I

5'-ACTCGAGAACGCAGAATTTTCGAGTTATT

c) Oligos for adding restriction sites to pACT upstream of GAL-4 AD to produce MCS-III

Sequence A5 [SEQ ID NO: 77]:

Spe I Sph I BssH2

5'-ATATGACTAGTGGCATGCGCGCCAATTTTAATCAAAGTGGG

Sequence A6 [SEQ ID NO: 78]:

Spe I ApaI SgrA I

5'-ATATGACTAGTGGGCCCCACCGGTGGCGGTACCCAATTCGACCTT

SEQUENCE LISTING

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Hua, Shaobing B.
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Lin, Yuhuei

<120> ASSEMBLY AND SCREENING OF HIGHLY COMPLEX AND FULLY
HUMAN ANTIBODY REPERTOIRE IN YEAST

<130> 25636-705 Seq Listing

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<210> 3

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<400> 3
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<210> 9
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<210> 10
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<210> 11
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<210> 12
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34

<210> 13

<211> 34

<212> DNA

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<400> 13

attacctcgt atagcataca ttatatgaag ttat

34

<210> 14

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46

<210> 15

<211> 45

<212> DNA

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45

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<212> DNA

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46

<210> 17

<211> 45

<212> DNA

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<211> 47

<212> DNA

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<223> Description of Artificial Sequence: PCR primer

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47

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<210> 22

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<210> 23

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<212> DNA

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<211> 38

<212> DNA

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48

<210> 28

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<212> DNA

<213> Artificial Sequence

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42

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<223> Description of Artificial Sequence: PCR primer

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aattttatgc tgactcagcc cc 82

<210> 38

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<223> Description of Artificial Sequence: PCR primer

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taggacggts ascttggtcc 80

<210> 39

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 39

gagatggtgc acgatgcaca gttgaagtga acttgcgagg tttttcagta tctacgattc 60
gaggacggtc agctgggtgc 80

<210> 40

<211> 87

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 40

gagatggtgc acgatgcaca gttgaagtga acttgcgagg tttttcagta tctacgattc 60
ttatgaacat tctgcagggg cmactgt 87

<210> 41

<211> 87

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 41

gagatggtgc acgatgcaca gttgaagtga acttgcgggg tttttcagta tctacgattc 60
ttaagagcat tctgcagggg ccactgt 87

<210> 42

<211> 83

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 42

ccaccaaacc caaaaaaaga gatctgtatg gcttacccat acgatgttcc agattacgct 60
gacatccrgd tgacccagtc tcc 83

<210> 43

<211> 83

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 43

ccaccaaacc caaaaaaaga gatctgtatg gcttacccat acgatgttcc agattacgct 60
gaaattgtrw tgacrcagtc tcc 83

<210> 44

<211> 83

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 44

ccaccaaacc caaaaaaaga gatctgtatg gcttacccat acgatgttcc agattacgct 60
gatattgtgm tgacbcagwc tcc 83

<210> 45

<211> 82

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 45

ccaccaaacc caaaaaaaga gatctgtatg gcttacccat acgatgttcc agattacgct 60
gaaacgacac tcacgcagtc tc 82

<210> 46

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 46

gagatggtgc acgatgcaca gttgaagtga acttgcgggg tttttcagta tctacgattc 60
tttgatttcc accttggtcc 80

<210> 47

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 47

gagatggtgc acgatgcaca gttgaagtga acttgcgggg tttttcagta tctacgattc 60
tttgatctcc ascttggtcc 80

<210> 48

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 48

gagatggtgc acgatgcaca gttgaagtga acttgcgggg tttttcagta tctacgattc 60
tttgatatcc acttggtcc 80

<210> 49

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 49

gagatggtgc acgatgcaca gttgaagtga acttgcgggg tttttcagta tctacgattc 60
ttaaattctcc agtcgtgtcc 80

<210> 50

<211> 84

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 50

gagatggtgc acgatgcaca gttgaagtga acttgcgggg tttttcagta tctacgattc 60
ctagcactct cccctgttga agct 84

<210> 51

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 51

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaagggtcga attgggtacc 60
gcccagtctg tsbtgacgca gccgcc 86

<210> 52

<211> 85

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 52

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaagggtcga attgggtacc 60
gcctcctatg wgctgacwca gccac 85

<210> 53

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 53

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gcctcctatg agctgayrca gcyacc 86

<210> 54

<211> 83

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 54

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gcccagcctg tgetgactca ryc 83

<210> 55

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 55

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gcccagdctg tgggtgacyca ggagcc 86

<210> 56

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 56

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gcccagccwg kgctgactca gccmcc 86

<210> 57

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 57

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gcctcctctg agctgastca ggascc 86

<210> 58

<211> 84

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 58

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gcccagtctg yyctgaytca gcct 84

<210> 59

<211> 85

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 59

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gcccaatttta tgctgactca gcccc 85

<210> 60

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 60

gagatggtgc acgatgcaca gttgaagtga acttgcgagg tttttcagta tctacgattc 60
taggacggtg ascttggtcc 80

<210> 61

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 61

gagatggtgc acgatgcaca gttgaagtga acttgcgggg tttttcagta tctacgattc 60
gaggacggtc agctgggtgc 80

<210> 62

<211> 84

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 62

gttagtgaaa gtgaaggaca atgagctatc agcaatattc ccactttgat taaaattggc 60
tgaacattct gcaggggcma ctgt 84

<210> 63

<211> 84

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 63

gttagtgaaa gtgaaggaca atgagctatc agcaatattc ccactttgat taaaattggc 60
agagcattct gcagggggcca ctgt 84

<210> 64

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 64

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gccgacatcc rgdtgaccca gtctcc 86

<210> 65

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 65

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gccgaaattg trwtgacrca gtctcc 86

<210> 66

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 66

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gccgatattg tgmtgacbca gwctcc 86

<210> 67

<211> 85

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 67

gataaagcgg aattaattcc cgagcctcca aaaaagaaga gaaaggtcga attgggtacc 60
gccgaaacga cactcacgca gtctc 85

<210> 68

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 68

gttagtgaaa gtgaaggaca atgagctatc agcaatattc ccactttgat taaaattggc 60
tttgatttcc accttggtcc 80

<210> 69

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 69

gtagtgaaa gtgaaggaca atgagctatc agcaatattc ccactttgat taaaattggc 60
tttgatctcc ascttggtcc 80

<210> 70

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 70

gtagtgaaa gtgaaggaca atgagctatc agcaatattc ccactttgat taaaattggc 60
tttgatatcc actttggtcc 80

<210> 71

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 71

gtagtgaaa gtgaaggaca atgagctatc agcaatattc ccactttgat taaaattggc 60
tttaatctcc agtcgtgtcc 80

<210> 72

<211> 81

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 72

gtagtgaaa gtgaaggaca atgagctatc agcaatattc ccactttgat taaaattggc 60
gcactctccc ctgttgaagc t 81

<210> 73

<211> 20

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Oligo for
mutation

<400> 73
gatccgcggc agctgtcgac

20

<210> 74
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Oligo for
mutation

<400> 74
gtacgtcgac agctgccgcg

20

<210> 75
<211> 28
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer

<400> 75
actcgagctt ctaattcttc caacatac

28

<210> 76
<211> 29
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer

<400> 76
actcgagaac gcagaatttt cgagttatt

29

<210> 77
<211> 41
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer

<400> 77
atatgactag tggcatgcgc gccaatTTta atcaaagtgg g

41

<210> 78

<211> 44

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PCR primer

<400> 78

atatgactag tgggcccacc ggtggcggtta cccaattcga cctt

44

<210> 79

<211> 24

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Semi-rigid
linker

<400> 79

Pro Gln Pro Gln Pro Lys Pro Gln Pro Gln Pro Gln Pro Gln Pro Lys
1 5 10 15

Pro Gln Pro Lys Pro Glu Pro Glu
20